Hines 10/026,914

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     (FILE 'HOME' ENTERED AT 16:01:45 ON 19 AUG 2003)

     FILE 'MEDLINE, HCAPLUS, BIOSIS, EMBASE, SCISEARCH, AGRICOLA' ENTERED AT
     16:02:08 ON 19 AUG 2003
L1           19 S LINHART B?/AU
L2         2280 S KRAFT D?/AU
L3         1250 S VALENTA R?/AU
L4         2766 S L1-L3
L5       112062 S ALLERGEN#
L6         1484 S L4 AND L5
L7           11 S L6 AND HYBRID#
L8          236 S (HYBRID# OR FUSION# OR CHIMER? OR CHIMAER?) (5A)ALLERGEN#
L9           25 S L8 AND GRASS?
L10          87 S PHL(A)P(A)I
L11           6 S L10 AND L8
L12           3 S PHL(A)P1
L13         223 S PHL(A)P(A)1
L14           3 S L13 AND L8
L15           1 S PHL(A)PI
L16           5 S L8(5A)TWO
L17           1 S L8(5A)THREE
L18          25 S L8 AND (REDUC? OR DECREAS? OR LESSEN? OR DIMINISH?)(3A)ALLER
L19          66 S L7 OR L9 OR L11 OR L12 OR L14-L18
L20         139 S FUSION(3A)PROTEIN#(5A)ALLERGEN?
L21           3 S L20 AND (REDUC? OR DECREAS? OR LESSEN? OR DIMINISH?)(3A)ALLE
L22           1 S ZIPPER? AND (REDUC? OR DECREAS? OR LESSEN? OR DIMINISH?)(3A)A
L23          67 S L19 OR L21 OR L22
L24          36 DUP REM L23 (31 DUPLICATES REMOVED)

=> d ibib abs l24 1-36

L24  ANSWER 1 OF 36  HCAPLUS  COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER:         2002:696098  HCAPLUS
DOCUMENT NUMBER:          137:231352
TITLE:                    Recombinant hybrid vespid venom
                          allergen constructs with reduced
                          allergenicity that retain immunogenicity of
                          the natural allergen
INVENTOR(S):              King, Te Piao; Spangfort, Michael Dho
PATENT ASSIGNEE(S):       The Rockefeller University, USA; Alk Abello A/S
SOURCE:                   PCT Int. Appl., 222 pp.
                          CODEN: PIXXD2
DOCUMENT TYPE:            Patent
LANGUAGE:                 English
FAMILY ACC. NUM. COUNT:   1
PATENT INFORMATION:

     PATENT NO.        KIND  DATE          APPLICATION NO.  DATE
     ---------------   ----  --------      ---------------  --------
     WO 2002070665      A2   20020912      WO 2002-US6765   20020304
         W:  AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
             CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
             GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
             LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH,
             PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ,
             UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU,
             TJ, TM
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Search completed by David Schreiber 308-4292

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

US 2003039660 A1 20030227 US 2002-91135 20020304
PRIORITY APPLN. INFO.: US 2001-272818P P 20010302

AB Disclosed are recombinant hybrid proteins having at least one antigenic peptide sequence introduced into a scaffold protein that retain a native conformation. Also disclosed are recombinant nucleic acids and vectors encoding the hybrid proteins. The hybrid proteins retain immunogenicity but exhibit **reduced allergenicity**. The hybrid proteins are therefore particularly useful for therapeutic treatment of allergy. The hybrid proteins are vespid venom antigen 5 proteins derived from genus Vespula or Polistes, e.g. Vespula vulgaris or Polistes annularis.

L24 ANSWER 2 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2002:517929 HCAPLUS
DOCUMENT NUMBER: 137:77892
TITLE: **Hybrid allergens** for therapy and diagnosis
INVENTOR(S): **Linhart, Birgit; Kraft, Dietrich; Valenta, Rudolf**
PATENT ASSIGNEE(S): Shan-Beteiligungsgesellschaft m.b.H., Austria
SOURCE: Eur. Pat. Appl., 23 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1221317	A1	20020710	EP 2000-128660	20001228
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
EP 1219301	A1	20020703	EP 2001-130292	20011220
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
US 2002173625	A1	20021121	US 2001-26914	20011227

PRIORITY APPLN. INFO.: EP 2000-128660 A 20001228

AB The authors disclose the construction and characterization of **hybrid** polypeptides comprising at least two different allergenic proteins or allergenic fragments. In one example, the **hybrid allergens** are constructed from the group 2 and 6 **allergens** of timothy **grass**. The **hybrid allergen** was shown to retain reactivity with serum antibodies of individuals with type I allergy.

REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L24 ANSWER 3 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2002:503332 HCAPLUS
DOCUMENT NUMBER: 137:62158
TITLE: Vaccines comprising chimeric protein of at least two different allergenic proteins for treating type I allergy
INVENTOR(S): **Linhart, Birgit; Kraft, Dietrich; Valenta, Rudolf**
PATENT ASSIGNEE(S): Shan-Beteiligungsgesellschaft M.B.H., Austria

SOURCE: Eur. Pat. Appl., 30 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 1219301	A1	20020703	EP 2001-130292	20011220
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
EP 1221317	A1	20020710	EP 2000-128660	20001228
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.: EP 2000-128660 A 20001228

AB **Hybrid** polypeptides comprising at least two different allergenic proteins or fragments thereof wherein each fragment consists of at least eight consecutive amino acids of the resp. allergenic protein are disclosed. The **hybrid** polypeptides and polynucleotides coding therefor can be used as pharmaceutical compns., in particular as vaccines. Thus, **hybrid** polypeptides comprising .gtoreq.2 timothy **grass** pollen **allergens** selecting from **Phl p 1, Phl p** 2, Phl p 5 and Phl p 6 were prepd. for inducing immune tolerance toward timothy **grass** pollen.

REFERENCE COUNT: 6 THERE ARE 6 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L24 ANSWER 4 OF 36 MEDLINE on STN DUPLICATE 1
ACCESSION NUMBER: 2002406202 MEDLINE
DOCUMENT NUMBER: 22148743 PubMed ID: 12154002
TITLE: Combination vaccines for the treatment of grass pollen allergy consisting of genetically engineered **hybrid** molecules with increased immunogenicity.
AUTHOR: **Linhart Birgit**; Jahn-Schmid Beatrice; Verdino Petra; Keller Walter; Ebner Christof; **Kraft Dietrich**; **Valenta Rudolf**
CORPORATE SOURCE: Department of Pathophysiology, Vienna General Hospital, AKH, University of Vienna, Austria.
SOURCE: FASEB JOURNAL, (2002 Aug) 16 (10) 1301-3.
Journal code: 8804484. ISSN: 1530-6860.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200209
ENTRY DATE: Entered STN: 20020806
Last Updated on STN: 20030105
Entered Medline: 20020903

AB Most of the 400 million grass pollen-allergic patients worldwide are co-sensitized to several unrelated grass pollen **allergens**. Based on frequent co-sensitization patterns determined in 200 grass pollen-allergic patients, three recombinant **hybrid** molecules were developed by polymerase chain reaction-based mending of cDNAs coding for the major timothy grass pollen **allergens** (Phl p 1, Phl p 2, Phl p 5, Phl p 6) for vaccination against grass pollen allergy. The **hybrids** rP2-P6, rP6-P2, and rP5-P1 contained most of the epitopes of natural grass pollen extract and induced stronger lymphoproliferative

responses in cultured mononuclear cells of grass pollen-allergic patients than did equimolar mixtures of the individual **allergens**. Immunization of mice with the **hybrids** yielded higher antibody titers than did immunization with the individual **allergen** components or grass pollen extract, which suggests that the individual components of the **hybrids** can serve as molecular scaffolds for each other to enhance their immunogenicity. Antibodies induced with the **hybrids** in mice inhibited the binding of grass pollen-allergic patients' immunoglobulin E to each of the individual **allergens** and grass pollen extract and may thus represent protective antibodies. The principle of increasing the immunogenicity of antigens by engineering **hybrids** thereof may be applied not only for the treatment of polysensitized allergic patients but also for general vaccine development.

L24 ANSWER 5 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2002:557372 HCAPLUS
DOCUMENT NUMBER: 138:23198
TITLE: Genetic engineering of allergens: future therapeutic products
AUTHOR(S): Ferreira, Fatima; Wallner, Michael; Breiteneder, Heimo; Hartl, Arnulf; Thalhamer, Josef; Ebner, Christof
CORPORATE SOURCE: Institute of Genetics, University of Salzburg, Salzburg, Austria
SOURCE: International Archives of Allergy and Immunology (2002), 128(3), 171-178
CODEN: IAAIEG; ISSN: 1018-2438
PUBLISHER: S. Karger AG
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English
AB A review. Genetic engineering of allergens for specific immunotherapy should aim at the prodn. of modified mols. with reduced IgE-binding epitopes (hypoallergens), while preserving structural motifs necessary for T cell recognition (T cell epitopes) and for induction of IgG antibodies reactive with the natural allergen (blocking antibodies). Common approaches for engineering of hypoallergens usually require knowledge of T and B cell epitopes and involve changing specific base pairs (mutated gene), introduction of a new piece of DNA into the existing DNA mol. (chimeric or hybrid gene), and deletions (truncated gene or fragments). DNA family shuffling has the advantage that it does not require a priori knowledge of structural and functional properties for efficient generation of hypoallergens. The combination of the hypoallergen concept with the Th1-inducing genetic immunization approach might be an attractive alternative for protein-based immunotherapy.
REFERENCE COUNT: 66 THERE ARE 66 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L24 ANSWER 6 OF 36 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
ACCESSION NUMBER: 2002:589912 SCISEARCH
THE GENUINE ARTICLE: 570WR
TITLE: Combination vaccines for the treatment of **grass** pollen allergy consisting of genetically engineered **hybrid** molecules with increased immunogenicity
AUTHOR: **Linhart B**; Jahn-Schmid B; Verdino P; Keller W; Ebner C; **Kraft D**; **Valenta R (Reprint)**
CORPORATE SOURCE: Univ Vienna, Sch Med, AKH, Mol Immunopathol Grp, Vienna Gen Hosp, Dept Pathophysiol, Waehringer Guertel 18-20, A-1090 Vienna, Austria (Reprint); Univ Vienna, Sch Med, AKH, Mol Immunopathol Grp, Vienna Gen Hosp, Dept

Search completed by David Schreiber 308-4292

Pathophysiol; A-1090 Vienna, Austria; Karl Franzens Univ Graz, Inst Chem, Graz, Austria
COUNTRY OF AUTHOR: Austria
SOURCE: FASEB JOURNAL, (JUN 2002) Vol. 16, No. 8, pp. U100-U124. Publisher: FEDERATION AMER SOC EXP BIOL, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3998 USA. ISSN: 0892-6638.
DOCUMENT TYPE: Article; Journal
LANGUAGE: English
REFERENCE COUNT: 59

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Most of the 400 million **grass** pollen-allergic patients worldwide are co-sensitized to several unrelated **grass** pollen **allergens**. Based on frequent co-sensitization patterns determined in 200 **grass** pollen-allergic patients, three recombinant **hybrid** molecules were developed by polymerase chain reaction-based mending of cDNAs coding for the major timothy **grass** pollen **allergens** (Phl p 1, Phl p 2, Phl p 5, Phl p 6) for vaccination against **grass** pollen allergy. The **hybrids** rP2-P6, rP6-P2, and rP5-P1 contained most of the epitopes of natural **grass** pollen extract and induced stronger lymphoproliferative responses in cultured mononuclear cells of **grass** pollen-allergic patients than did equimolar mixtures of the individual **allergens**. Immunization of mice with the **hybrids** yielded higher antibody titers than did immunization with the individual **allergen** components or **grass** pollen extract, which suggests that the individual components of the **hybrids** can serve as molecular scaffolds for each other to enhance their immunogenicity. Antibodies induced with the **hybrids** in mice inhibited the binding of **grass** pollen-allergic patients' immunoglobulin E to each of the individual **allergens** and **grass** pollen extract and may thus represent protective antibodies. The principle of increasing the immunogenicity of antigens by engineering **hybrids** thereof may be applied not only for the treatment of polysensitized allergic patients but also for general vaccine development.

L24 ANSWER 7 OF 36 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 2001448606 MEDLINE
DOCUMENT NUMBER: 21240667 PubMed ID: 11342623
TITLE: Recombinant **allergens** with **reduced allergenicity** but retaining immunogenicity of the natural **allergens**: **hybrids** of yellow jacket and paper wasp venom allergen antigen 5s.
AUTHOR: King T P; Jim S Y; Monsalve R I; Kagey-Sobotka A; Lichtenstein L M; Spangfort M D
CORPORATE SOURCE: Rockefeller University, New York, NY 10021, USA.. kingtp@mail.rockefeller.edu
CONTRACT NUMBER: AI-08270 (NIAID)
AI-17021 (NIAID)
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 May 15) 166 (10) 6057-65. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200108
ENTRY DATE: Entered STN: 20010813
Last Updated on STN: 20010813
Entered Medline: 20010809

Search completed by David Schreiber 308-4292

AB The homologous venom allergen Ag 5s from the yellow jacket (Vespula vulgaris) and paper wasp (Polistes annularis) have 59% sequence identity of their respective 204 and 205 amino acid residues, and they have low degrees of antigenic cross-reactivity in insect allergic patients and in animal models. Hybrids containing different segments of these two vespid Ag 5s were expressed in yeast. Circular dichroism spectroscopy suggests the hybrids to have the secondary structure of natural Ag 5. Inhibition ELISA with human and murine Abs suggests the hybrids to have the discontinuous B cell epitopes of the natural Ag 5 but with an altered epitope density. The hybrids were immunogenic in mice for B and T cell responses to both Ag 5s. The N-terminal region of Ag 5 was found to contain its dominant B cell epitope(s). Hybrids containing 10-49 residues of yellow jacket Ag 5 showed 100- to 3000-fold **reduction** in **allergenicity** when tested by histamine release assay with basophils of yellow jacket-sensitive patients. Our findings suggest that hybrids represent a useful approach to map the discontinuous B cell epitope-containing regions of proteins. They also suggest that Ag 5 hybrids may be useful immunotherapeutic reagents in man.

L24 ANSWER 8 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2002:13053 HCAPLUS
DOCUMENT NUMBER: 137:19252
TITLE: Measurement of IgE antibodies against purified grass-pollen allergens (Phl p 1, 2, 3, 4, 5, 6, 7, 11, and 12) in sera of patients allergic to grass pollen
AUTHOR(S): Rossi, R. E.; Monasterolo, G.; Monasterolo, S.
CORPORATE SOURCE: Servizio di Immunoallergologia Ospedale S.S.Annunziata, Savigliano, Italy
SOURCE: Allergy (Copenhagen, Denmark) (2001), 56(12), 1180-1183
CODEN: LLRGDY; ISSN: 0105-4538
PUBLISHER: Munksgaard International Publishers Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Current allergy diagnosis is performed with allergen exts. which contain a variety of allergenic and nonallergenic components. The availability of highly purified and well-characterized allergen mols. seems to be an advantage of component-based diagnosis. With the immunoenzymic CAP FEIA System, we measured specific IgE levels to the recombinant allergens rPhl p 1, rPhl p 2, rPhl p 5, rPhl p 6, rPhl p 7, rPhl p 11, rPhl p 12, and native Phl p 4 in 77 sera of patients allergic to grass pollen, in order to evaluate the IgE-binding frequency to these purified grass-pollen allergens and their relationship to rBet v 4, rBet v 2, and other allergens. The frequency of sensitization was as follows: rPhl p 1 = 93.5%; rPhl p 2 = 67.5%; rPhl p 5 = 72.7%; rPhl p 6 = 68.8%; rPhl p 7 = 7.8%; rPhl p 11 = 53.2%; rPhl p 12 = 35.1%; and native Phl p 4 = 88.3%. As expected, rPhl p 7 and rPhl p 12 had a very good correlation (Spearman's r) with Bet v 4 and rBet v 2. Good correlations of rPhl p 12. with papain, latex, and bromelain were found. Highly variable individual sensitization patterns were obsd. A new clin. approach has allowed the detn. of specific allergograms for the different patients and may therefore be of great importance for more specific diagnosis. The use of component-resolved diagnostics may be useful to evaluate the allergen content of an ext. for immunotherapy by monitoring patient's IgE and IgG directed to relevant allergens.
REFERENCE COUNT: 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L24 ANSWER 9 OF 36 MEDLINE on STN DUPLICATE 3

Search completed by David Schreiber 308-4292

ACCESSION NUMBER: 2001139800 MEDLINE
DOCUMENT NUMBER: 20581159 PubMed ID: 11145673
TITLE: Vaccination with **allergen**-IL-18 **fusion** DNA protects against, and reverses established, airway hyperreactivity in a murine asthma model.
AUTHOR: Maecker H T; Hansen G; Walter D M; DeKruyff R H; Levy S; Umetsu D T
CORPORATE SOURCE: Division of Oncology, Department of Medicine, and the Division of Immunology and Transplantation Biology, Department of Pediatrics, Stanford University Medical Center, Stanford, CA 94305, USA.
CONTRACT NUMBER: AI-07290-14 (NIAID)
AI37219 (NIAID)
AI45900 (NIAID)
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Jan 15) 166 (2) 959-65.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200103
ENTRY DATE: Entered STN: 20010404
Last Updated on STN: 20010404
Entered Medline: 20010308

AB Vaccination with naked DNA encoding a specific allergen has been shown previously to prevent, but not reverse, the development of allergen-induced airway hyperresponsiveness (AHR). To enhance the effectiveness of DNA vaccine therapies and make possible the treatment of established AHR, we developed a DNA vaccination plasmid containing OVA cDNA fused to IL-18 cDNA. Vaccination of naive mice either with this fusion DNA construct or with an OVA cDNA-containing plasmid protected the mice from the subsequent induction of AHR. Protection from AHR correlated with increased IFN-gamma production and reduced OVA-specific IgE production. The protection appeared to be mediated by IFN-gamma and CD8(+) cells because treatment of mice with neutralizing anti-IFN-gamma mAb or with depleting anti-CD8 mAb abolished the protective effect. Moreover, vaccination of mice with preexisting AHR with the OVA-IL-18 fusion DNA, but not with the OVA cDNA plasmid, reversed established AHR, **reduced allergen**-specific IL-4, and increased allergen-specific IFN-gamma production. Thus, combining IL-18 cDNA with OVA cDNA resulted in a vaccine construct that protected against the development of AHR, and that was unique among cDNA constructs in its capacity to reverse established AHR.

L24 ANSWER 10 OF 36 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:495323 BIOSIS
DOCUMENT NUMBER: PREV200100495323
TITLE: Recombinant **hybrid allergens** for the treatment of **grass** pollen allergy.
AUTHOR(S): **Linhart, B. (1); Kraft, D. (1); Valenta, R. (1)**
CORPORATE SOURCE: (1) Inst. f. Pathophysiology, University of Vienna, Vienna Austria
SOURCE: Allergy (Copenhagen), (2001) Vol. 56, No. Supplement 68, pp. 82. print.
Meeting Info.: XXth Congress of the European Academy of Allergology and Clinical Immunology Berlin, Germany May 09-13, 2001
ISSN: 0105-4538.

Search completed by David Schreiber 308-4292

DOCUMENT TYPE: Conference
LANGUAGE: English
SUMMARY LANGUAGE: English

L24 ANSWER 11 OF 36 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:506770 BIOSIS
DOCUMENT NUMBER: PREV200100506770
TITLE: A novel type of a vaccine for bee venom allergy obtained by gene **fusion** of the **two** major **allergens**.
AUTHOR(S): Kussebi, F. (1); Rhyner, C. (1); Akdis, M. (1); Schmid, P. (1); Blaser, K. (1); Crameri, R. (1); Akdis, C. (1)
CORPORATE SOURCE: (1) Swiss Institute of Allergy and Asthma Research, (SIAF), Davos Switzerland
SOURCE: Allergy (Copenhagen), (2001) Vol. 56, No. Supplement 68, pp. 13. print.
Meeting Info.: XXth Congress of the European Academy of Allergology and Clinical Immunology Berlin, Germany May 09-13, 2001
ISSN: 0105-4538.
DOCUMENT TYPE: Conference
LANGUAGE: English
SUMMARY LANGUAGE: English

L24 ANSWER 12 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 2000:163611 HCAPLUS
DOCUMENT NUMBER: 133:162982
TITLE: Phleum pratense-specific T cells of allergic rhinitis patients display a broader recognition pattern than Phleum pratense-specific serum immunoglobulin E
AUTHOR(S): Van Neerven, R. J. J.; Arnved, J.; Ipsen, H.
CORPORATE SOURCE: ALK-Abello, Horsholm, Den.
SOURCE: Clinical and Experimental Allergy (2000), 30(2), 242-254
CODEN: CLEAEN; ISSN: 0954-7894
PUBLISHER: Blackwell Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The role of allergen-specific CD4+ T lymphocytes in the pathophysiol. of atopic disease is well established. Previous studies on allergen-specific T-cell responses have focused on the recognition of single major allergens to identify T-cell epitopes. However, it is not clear whether immune responses to allergen exts. are exclusively targeted at major allergens or whether addnl. proteins are recognized. Here the authors describe the P. pratense-specific IgE and T-cell responses of 6 allergic rhinitis patients. Reactivity was measured to size-sepd. fractions of a P. pratense ext. as well as to the purified major allergens Phl p 1, Phl p 2/3, and Phl p 5. The specificity of the patients' serum IgE, measured in a fluid phase assay, was restricted to 1 or 2 of the major allergens. Even though the majority of the patients had IgE antibodies reactive with a single major allergen, one patient reacted with both Phl p 5 and with Phl p 2/3. Anal. of the T-cell repertoire with P. pratense-specific T-cell lines (TCLs) and CD4+ T-cell clones (TCCs) revealed that at least 6 different proteins were recognized, including the 3 major allergens, most notably Phl p 5. Simultaneous prodn. of IL-5 and interferon (IFN)-.gamma. was detected in supernatants of the TCLs stimulated with P. pratense ext. and the major allergens. Thus, allergic rhinitis patients have a large pool of circulating allergen-specific CD4+ T cells that recognize many different proteins in an allergenic ext., whereas only a small no. of

these proteins are recognized by serum IgE.
REFERENCE COUNT: 61 THERE ARE 61 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L24 ANSWER 13 OF 36 MEDLINE on STN DUPLICATE 4
ACCESSION NUMBER: 2000195492 MEDLINE
DOCUMENT NUMBER: 20195492 PubMed ID: 10729775
TITLE: Molecular breeding of allergy vaccines and antiallergic cytokines.
AUTHOR: Punnonen J
CORPORATE SOURCE: Maxygen, Redwood City, CA 94063, USA.
SOURCE: INTERNATIONAL ARCHIVES OF ALLERGY AND IMMUNOLOGY, (2000 Mar) 121 (3) 173-82. Ref: 85
Journal code: 9211652. ISSN: 1018-2438.
PUB. COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000620

AB Molecular breeding, also called DNA shuffling, is a technology that enables the generation of large libraries of novel genes and vectors, from which improved variants can be selected based on functional properties. In a common format, it involves recursive recombination and mutation, performed by random fragmentation of related DNA sequences, followed by reassembly of the fragments in a self-priming polymerase chain reaction. As in natural evolution, the technique takes advantage of crossovers, deletions, insertions, inversions and point mutations of genes to generate large pools of related sequences. Molecular breeding can be used to generate improved variants of proteins used as therapeutics, such as vaccine antigens, growth factors and immunomodulatory molecules. Moreover, the technology can be applied to evolve entire viruses or vectors, including DNA vaccines. Cytokines downregulating allergic immune responses and allergens are attractive targets for evolution by molecular breeding. This review describes approaches to generate **chimeric allergens** with T cell epitopes from multiple **allergen** homologues, while **reducing** the recognition by preexisting IgE. In addition, the results and applications of molecular breeding in the evolution of improved antiallergic cytokines are discussed.

L24 ANSWER 14 OF 36 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
ACCESSION NUMBER: 1999:468170 SCISEARCH
THE GENUINE ARTICLE: 205NV
TITLE: Induction of antibody responses to new B cell epitopes indicates vaccination character of allergen immunotherapy
AUTHOR: Ball T; Sperr W R; Valent P; Lidholm J; Spitzauer S; Ebner C; Kraft D; Valenta R (Reprint)
CORPORATE SOURCE: UNIV VIENNA, SCH MED, GEN HOSP, INST GEN & EXPT PATHOL, MOL IMMUNOPATHOL GRP, A-1090 VIENNA, AUSTRIA (Reprint); UNIV VIENNA, SCH MED, GEN HOSP, INST GEN & EXPT PATHOL, MOL IMMUNOPATHOL GRP, A-1090 VIENNA, AUSTRIA; UNIV VIENNA, GEN HOSP, DEPT INTERNAL MED 1, DIV HEMATOL & HEMOSTASEOL, A-1090 VIENNA, AUSTRIA; PHARMACIA & UPJOHN INC, AB DIAGNOST, UPPSALA, SWEDEN; UNIV VIENNA, DEPT MED & CHEM

Search completed by David Schreiber 308-4292

LAB DIAGNOST, GEN HOSP, A-1090 VIENNA, AUSTRIA
COUNTRY OF AUTHOR: AUSTRIA; SWEDEN
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (JUN 1999) Vol. 29, No. 6, pp. 2026-2036.
Publisher: WILEY-V C H VERLAG GMBH, MUHLENSTRASSE 33-34, D-13187 BERLIN, GERMANY.
ISSN: 0014-2980.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 54

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Whether the modulation of antibody responses can contribute to the improvement of clinical symptoms in patients receiving allergen immunotherapy represents a controversial issue. We have used purified [seven recombinant (r) and one natural] timothy grass pollen allergens as well as recombinant B cell epitope-containing fragments of the major timothy grass pollen allergen, Phl p 1, to investigate humoral immune responses in eight allergic patients receiving grass pollen-specific immunotherapy. We found that the administration of aluminium hydroxide-adsorbed grass pollen extract induced complex changes in allergen/epitope-specific ;antibody responses: increases in IgG subclass (IgG1, IgG2, IgG4) responses against allergens recognized before the therapy were observed. All eight patients started to mount IgE and IgG4 responses to continuous Phl p 1 epitopes not recognized before the therapy and a de novo induction of IgE antibodies against new allergens was found in one patient. Evidence for a protective role of IgG antibodies specific for continuous Phl p 1 epitopes was provided by the demonstration that preincubation of rPhl p 1 with human serum containing therapy-induced **Phl pi**-specific IgG inhibited rPhl p 1-induced histamine release from basophils of a grass pollen-allergic patient. Our finding that immunotherapy induced antibody responses against previously not recognized B cell epitopes indicates the vaccination character of this treatment. The fact that patients started to mount de novo IgE as well as protective IgG responses against epitopes may explain the unpredictability of specific immunotherapy performed with allergen extracts and emphasizes the need for novel forms of component-resolved immunolherapy.

L24 ANSWER 15 OF 36 MEDLINE on STN DUPLICATE 5
ACCESSION NUMBER: 2000059392 MEDLINE
DOCUMENT NUMBER: 20059392 PubMed ID: 10590258
TITLE: CD4(+) T cells induced by virus-like particles expressing a major T cell epitope down-regulate IL-5 production in an ongoing immune response to Der p 1 independently of IFN-gamma production.
AUTHOR: Hirschberg S; Layton G T; Harris S J; Savage N; Dallman M J; Lamb J R
CORPORATE SOURCE: Department of Biology, Imperial College of Science, London SW7 2AZ, UK.
SOURCE: INTERNATIONAL IMMUNOLOGY, (1999 Dec) 11 (12) 1927-34.
Journal code: 8916182. ISSN: 0953-8178.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200001
ENTRY DATE: Entered STN: 20000204
Last Updated on STN: 20000204
Entered Medline: 20000124

Search completed by David Schreiber 308-4292

AB It has been previously demonstrated that hybrid Ty virus-like particles (VLP) prime effective CD8(+) and CD4(+) T cell responses. In this study, we investigated the effect of treating mice with Ty VLP carrying the immunodominant epitope of Der p 1 after sensitizing them to the group 1 allergen of the house dust mite Dermatophagoides pteronyssinus (Der p 1), under conditions that induce T(h)2 immunity. We show that i.p. treatment with the **hybrid** VLP abrogated **allergen**-specific IL-5 production and **reduced allergen**-specific cell proliferation. This suppression of the response was mediated by CD4(+) T cells and was not accompanied by an increase in IFN-gamma production.

L24 ANSWER 16 OF 36 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V. on STN
ACCESSION NUMBER: 1999148709 EMBASE
TITLE: Lessons from the antibody recognition of the major Timothy grass pollen allergen **Phl p1**.
AUTHOR: Ball T.; Fuchs T.; Kraft D.; Valenta R.
CORPORATE SOURCE: Dr. R. Valenta, Molecular Immunopathology Group, Dept. General Exptl. Pathology AKH, University of Vienna, Waehringer Guertel 18-20, A-1090 Vienna, Austria. A5311daa@awiunil1.edvz.univie.ac.at
SOURCE: International Archives of Allergy and Immunology, (1999) 118/2-4 (208-209).
Refs: 7
ISSN: 1018-2438 CODEN: IAAIEG
COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Conference Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
LANGUAGE: English

L24 ANSWER 17 OF 36 MEDLINE on STN DUPLICATE 6
ACCESSION NUMBER: 97430885 MEDLINE
DOCUMENT NUMBER: 97430885 PubMed ID: 9284984
TITLE: Nonspecific binding of IgE to allergens.
AUTHOR: Jensen-Jarolim E; Vogel M; Zavazal V; Stadler B M
CORPORATE SOURCE: Institute of General and Experimental Pathology, Allgemeines Krankenhaus Wien, Austria.
SOURCE: ALLERGY, (1997 Aug) 52 (8) 844-52.
Journal code: 7804028. ISSN: 0105-4538.
PUB. COUNTRY: Denmark
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199710
ENTRY DATE: Entered STN: 19971105
Last Updated on STN: 19971105
Entered Medline: 19971023

AB Nonspecific IgE binding to allergens was observed in testing myeloma IgEs, namely, IgE-VL and IgE-PS, chimeric IgE (IgE-JW8), and the recombinant IgE Fc epsilon peptide CH1-CH4, in two different immunoassays. This binding was concentration-dependent but detectable only at higher IgE concentration. In RAST inhibition, IgE-**allergen** interactions could be **reduced** by using either matching or nonmatching allergens. In order to test whether the nonspecific binding of IgE to allergens was due to carbohydrate interaction, myeloma IgEs and the chimeric IgE were desialized with neuraminidase. Desialized samples were equally well recognized by xenogenic antibodies as native IgEs, but binding of IgE to Fc epsilon receptors on basophils was affected by the treatment, as shown in the histamine-release assay. Desialization of IgE affected also its binding capacity to **allergens** in RAST: binding

of **chimeric** IgE was reduced, but nonspecific binding of myeloma IgE-VL was enhanced. Hence, nonspecific allergen-IgE binding may be partly due to a lectin-like interaction, but may depend mostly on the tertiary structure of IgE. Thus, nonspecific IgE-allergen interactions might present a problem 1) at high IgE concentration, and 2) depend on the grade of sialization of IgE, which might affect its conformation. This may explain why patients with elevated total IgE levels often have multiple weak positive RASTs with non-cross-reactive allergens.

L24 ANSWER 18 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1996:467157 HCAPLUS
DOCUMENT NUMBER: 125:108885
TITLE: **Zipper** techniques for producing polypeptides with **reduced allergenicity**
INVENTOR(S): Bjoernvad, Mads Eskelund; Prentoe, Annette
PATENT ASSIGNEE(S): Novo Nordisk A/s, Den.
SOURCE: PCT Int. Appl., 84 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9616177	A1	19960530	WO 1995-DK463	19951123
W: AL, AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK				
RW: KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9539240	A1	19960617	AU 1995-39240	19951123
EP 793726	A1	19970910	EP 1995-936995	19951123
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, NL, PT, SE				
JP 10509324	T2	19980914	JP 1995-516467	19951123

PRIORITY APPLN. INFO.: DK 1994-1343 19941124
WO 1995-DK463 19951123

AB Disclosed is a method for the prepn. of polypeptides with **reduced allergenicity** by recombinant prepn. of a **fusion protein** contg. the polypeptides and **Zipper** domain(s), whereby the expressed polypeptide mols. self-oligomerize. The polypeptides can be produced in transgenic microorganisms such as yeast, bacteria, and filamentous fungi. Prepn. of dimeric Termamyl using the Leucine **Zipper** of GCN4 in transgenic Escherichia coli JM105. The dimeric Termamyl exhibits >50% of the wild type activity. Applications of Termamyl in cosmetic, pharmaceutical, agricultural industries, etc. are claimed.

L24 ANSWER 19 OF 36 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
DUPLICATE 7
ACCESSION NUMBER: 1996:406916 BIOSIS
DOCUMENT NUMBER: PREV199699129272
TITLE: Sequencing analysis of cDNA clones encoding the American cockroach Cr-PI allergens. Homology with insect hemolymph proteins.
AUTHOR(S): Wu, Chii H. (1); Lee, Mey F.; Liao, Sin C.; Luo, Shue F.
CORPORATE SOURCE: (1) Dep. Med. Res., Taichung Veterans Gen. Hosp., 160

Chung-Kang Road, Sect. 3, Taichung 40705 Taiwan
SOURCE: Journal of Biological Chemistry, (1996) Vol. 271, No. 30, pp. 17937-17943.
ISSN: 0021-9258.
DOCUMENT TYPE: Article
LANGUAGE: English

AB A previous article described the isolation of several lambda-gt22A cDNA clones expressing the American cockroach (Periplaneta americana) Cr-PI allergens recognized by both human atopic IgE antibodies and anti-Cr-PI monoclonal antibodies (Wu, C. H, Lee, M. F., and Liao, S. C. (1995) J. Allergy Clin. Immunol. 96, 352-859). This article presents the nucleotide and deduced amino acid sequences of two cDNA clones encoding major allergens of P. americana. Clones C12 and C20 encode proteins of 685 and 631 amino acids with two potential N-glycosylation sites each. The predicted molecular weights for C12 and C20 cloned proteins are 79,300 and 75,500 with isoelectric point values of 6.26 and 6.63, which are compatible with the determined sizes (M-r 78,000 and 72,000) and isoelectric point value (6.2) of the Cr-PI allergens of P. americana. A high degree of identity (69.1%), including several overlapped predicted central antigenic determinant residues, was found between **two allergens**. The anti-**fusion** protein antibody-based enzyme-linked immunosorbent assay was able to detect crude American cockroach extract, Cr-PI, recombinant proteins, and commercial cockroach extracts, which provides further evidence that two allergens share common antigen determinants. Recombinant allergens of clones C12 and C20 both showed 47.4% skin reactivities on 19 cockroach-sensitive asthmatic patients. Unexpectedly, although no sequence similarity was found to other known allergens, two aromatic amino acid-rich allergens were found to have a striking sequence identity to insect storage proteins (20.1-33.9%), insect juvenile hormone-suppressible proteins (30.9-36.4%), and arthropod hemocyanins (29.7-34.6%). Results suggested that two prominent allergens of P. americana are ancestrally related to these insect hemolymph proteins and represent a now group of proteins in the hemocyanin superfamily. These data will now facilitate epitope-mapping studies, and the recombinant allergens may be valuable for diagnostic and therapeutic purposes.

L24 ANSWER 20 OF 36 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V. on STN

ACCESSION NUMBER: 96076387 EMBASE
DOCUMENT NUMBER: 1996076387

TITLE: Expression of Zm13, a pollen specific maize protein, in Escherichia coli reveals IgE-binding capacity and allergenic potential.

AUTHOR: Heiss S.; Flicker S.; Hamilton D.A.; **Kraft D.**; Mascarenhas J.P.; **Valenta R.**
CORPORATE SOURCE: Inst. of General and Exp. Pathology, AKH, University of Vienna, Wahringer Gurtel 18-20,A-1090 Vienna, Austria

SOURCE: FEBS Letters, (1996) 381/3 (217-221).
ISSN: 0014-5793 CODEN: FEBLAL
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
052 Toxicology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Plant proteins belong to the most frequent elicitors of type I allergic symptoms in industrialized countries. Several relevant plant **allergens** have been found to be either specifically expressed or

highly upregulated in mature pollen. The cDNA coding for a pollen specific maize protein, Zm13, shows significant sequence homology with a number of pollen or anther specific proteins from monocot and dicot plants as well as with recently described **allergens** from olive and rye grass. To test whether the Zm13 protein might possess IgE-binding capacity, Zm13 was expressed in E. coli. The coding region of Zm13 was PCR amplified from a genomic clone and expressed as as a glutathione-S-transferase fusion protein. The recombinant Zm13 fusion protein bound a Zm13 specific rabbit antiserum and reacted with serum IgE from grass pollen allergic patients indicating that Zm13 and homologous proteins represent a family of conserved plant **allergens**.

L24 ANSWER 21 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1996:73365 HCAPLUS
DOCUMENT NUMBER: 124:115472
TITLE: Recombinant allergen, fragments thereof, corresponding recombinant DNA molecules, vectors and hosts containing the DNA molecules, diagnostic and therapeutic uses of said allergens and fragments
INVENTOR(S): Ball, Tanja; Vrtala, Susanne; Sperr, Wolfgang; Valent, Peter; Susani, Markus; Kraft, Dietrich; Valenta, Rudolf; Laffer, Sylvia
PATENT ASSIGNEE(S): Pharmacia AB, Swed.
SOURCE: PCT Int. Appl., 24 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9534578	A1	19951221	WO 1995-SE724	19950614
W: AU, CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
AU 9527598	A1	19960105	AU 1995-27598	19950614
EP 763059	A1	19970319	EP 1995-922855	19950614
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, NL, SE				
JP 10501417	T2	19980210	JP 1995-502050	19950614
US 6008340	A	19991228	US 1997-750419	19970114
US 2002052490	A1	20020502	US 2001-811672	20010320
US 6559120	B2	20030506		

PRIORITY APPLN. INFO.:
SE 1994-2089 A 19940614
WO 1995-SE724 W 19950614
US 1997-750419 A3 19970114
US 1999-303684 A3 19990503

AB A recombinant DNA mol. comprising a nucleotide sequence (I) which codes for a polypeptide displaying the antigenicity of one, two or more of the **Phl p I** epitope clones (28, 34, 41, 42, 43, 50, 52, 64, 80, 85, 86, 95, 97, 98, 103, 108, 109, 113, 114) with the amino acid sequences defined in figure 2 and preferably being derived from **grasses** or monocotyledonic plants, or a nucleotide sequence (II) which hybridizes with such a nucleotide sequence (I) under conditions of high stringency. Polypeptides displaying the antigenicity of one, two or more of the **Phl p I** epitope clones (28, 34, 41, 42, 43, 50, 52, 64, 80, 85, 86, 95, 97, 98, 103, 108, 109, 113, 114) with the amino acid sequences defined in figure 2. Recombinant expression vectors contg. the recombinant mol. and host cells transformed with the vector. Diagnostic methods based on utilizing the polypeptides in

immunoassays for humoral antibodies and cellular reactions.

L24 ANSWER 22 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1995:846664 HCAPLUS
DOCUMENT NUMBER: 123:254590
TITLE: A novel allergen from Kentucky bluegrass that has cross-reactive homologs in mono- and dicotyledonous plants
INVENTOR(S): Mohapatra, Shyam S.; Sehon, Alec H.
PATENT ASSIGNEE(S): University of Manitoba, Can.
SOURCE: PCT Int. Appl., 49 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9519437	A1	19950720	WO 1995-CA21	19950116
W: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MN, MW, MX, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SI, SK, TJ, TT, UA, US				
RW: KE, MW, SD, SZ, AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
CA 2181142	AA	19950720	CA 1995-2181142	19950116
AU 9514109	A1	19950801	AU 1995-14109	19950116
PRIORITY APPLN. INFO.:			US 1994-181383	19940114
			WO 1995-CA21	19950116

AB A widely-occurring plant allergen that may be a useful target for immunotherapy of allergy (no data) is described. Screening of a Kentucky Bluegrass pollen-.lambda.gt11 library with sera from patients allergic to **grass** pollen led to the identification of a partial cDNA clone, KBG51. Nucleotide sequence anal. of KBG51 indicated that the polypeptide encoded by this cDNA is different from that of the known recombinant **grass** pollen allergens. Using murine antiserum to a GST-KBG51 fusion protein, produced with aid of the pGEX-2T-1 expression system, two polypeptides of about 30 and 59 KDa in size, were detectable in SDS-PAGE immunoblot anal. of KBG pollen proteins. The reactivity of this antiserum, with a no. of polypeptides, which ranged in size from 29.5 to 115 kDa, from pollen exts. of several **grasses**, birch, ragweed and Parietaria and the hybridization of RT-PCR products from various pollens with radiolabeled KBG51-cDNA, demonstrated the cross-reactivity (CR) of this AL with other pollen ALS. Because of the broad CR, the protein(s) corresponding to KBG51 has been designated as CRAL51. Anal. by ELISA using sera of about 1000 individuals worldwide who were allergic to pollens demonstrated that individuals from a variety of geog. areas possessed IgE antibodies that recognized the GST-KBG51 fusion protein. On the basis of these findings, CRAL51 represents a member of a family of highly cross-reactive ALs in plant pollens.

L24 ANSWER 23 OF 36 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V. on STN
ACCESSION NUMBER: 94350767 EMBASE
DOCUMENT NUMBER: 1994350767
TITLE: Isolation of an immunodominant IgE hapten from an epitope expression cDNA library. Dissection of the allergic effector reaction.

Search completed by David Schreiber 308-4292

AUTHOR: Ball T.; Vrtala S.; Sperr W.R.; Valent P.; Susani M.; **Kraft D.**; **Valenta R.**
CORPORATE SOURCE: General/Experimental Pathology Inst., AKH, University of Vienna, Wahringergurtel 18-20,A-1090 Vienna, Austria
SOURCE: Journal of Biological Chemistry, (1994) 269/45 (28323-28328).
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English

AB An epitope expression cDNA library was constructed from the randomly fragmented cDNA coding for Phl p I, the major grass pollen **allergen**. Using IgE from allergic patients, epitope clones were isolated and immunodominant fragments were selected. Among three epitope clones coding for a similar region of Phl p I, one clone expressed a 15-amino-acid epitope which was target for IgE antibodies from approximately 30% of grass pollen allergic patients. According to the prevalence of grass pollen allergy, 22% of all allergic patients are expected to display IgE reactivity with this epitope. Although the purified recombinant epitope specifically bound IgE, it did not release histamine from basophiles of most grass pollen allergic patients and thus represents an IgE hapten. Immunodominant IgE haptens may be useful as therapeutic agents to saturate mast cell-bound IgE prior to **allergen** exposure and may represent candidates for a safe immunotherapy of allergic diseases by reducing anaphylactic side effects.

L24 ANSWER 24 OF 36 EMBASE
ACCESSION NUMBER: 94327920 EMBASE
DOCUMENT NUMBER: 1994327920
TITLE: Complementary DNA cloning of the major **allergen** Phl p I from timothy grass (Phleum pratense); recombinant Phl p I inhibits IgE binding to group I **allergens** from eight different grass species.
AUTHOR: Laffer S.; **Valenta R.**; Vrtala S.; Susani M.; Van Ree R.; **Kraft D.**; Scheiner O.; Duchene M.
CORPORATE SOURCE: General/Experimental Pathology Inst., AKH, Wahringergurtel 18-20,A-1090 Vienna, Austria
SOURCE: Journal of Allergy and Clinical Immunology, (1994) 94/4 (689-698).
ISSN: 0091-6749 CODEN: JACIBY
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Background: Grass pollens, such as pollen from timothy grass (Phleum pratense), represent a major cause of type 1 allergy. Objective: In this report we attempted to determine how cross-reactive allergenic components of grass pollens from different species can be represented by a minimum number of recombinant **allergens**. Methods: We isolated and sequenced a timothy grass pollen cDNA coding for the major **allergen** Phl p I. A recombinant Phl p I-.beta.- galactosidase fusion protein, which bound to IgE in 87% of patients with grass pollen allergy, was produced in Escherichia coli. Using recombinant Phl p V and

Phl p I, we defined representative patients' sera that bound to group I but not to group V **allergens**, as well as sera with reactivity against group I and group V **allergens**. IgE immunoblot inhibition studies were done with nitrocellulose-blotted pollen extracts from eight grass species with different geographic distribution. Results: Preadsorption of patients' sera with recombinant nonfusion Phl p I strongly reduced IgE binding to group I **allergens** from the eight grasses, showing extensive cross-reactivity between species. Conclusion: A single recombinant group I **allergen** contains many of the IgE epitopes of group I isoallergens from a number of different grass species.

L24 ANSWER 25 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1993:232280 HCAPLUS
DOCUMENT NUMBER: 118:232280
TITLE: A novel group of major **grass** pollen allergens and the genes encoding them
INVENTOR(S): Mohapatra, Shyam S.; Hill, Robert D.; Kisil, Fred T.; Sehon, Alec
PATENT ASSIGNEE(S): Can.
SOURCE: Can. Pat. Appl., 55 pp.
CODEN: CPXXEB
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
CA 2068694	AA	19930109	CA 1992-2068694	19920514
PRIORITY APPLN. INFO.:			US 1991-726937	19910708

AB Novel allergens of Kentucky bluegrass (Poa pratensis) are identified and cDNAs encoding them are cloned and expressed for use in diagnosis and treatment of the allergy. The cDNAs were cloned from an expression library in .lambda.gt11 by screening with serum from patients allergic to bluegrass pollen and by further screening of the bank with preliminary clones used to further screen the bank. Three clones were obtained and found to show only sporadic sequence similarities to other allergens. The cDNAs appear to be from members of a multigene family. Homologous genes were found in several other plant species. The cDNA was expressed in Escherichia coli and the immunol. active protein found in resolubilized inclusion bodies; subclones were used for epitope mapping.

L24 ANSWER 26 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1994:29262 HCAPLUS
DOCUMENT NUMBER: 120:29262
TITLE: Cloning, expression, and immunological characterization of recombinant Lolium perenne allergen Lol p II
AUTHOR(S): Sidoli, Alessandro; Tamborini, Elena; Giuntini, Ilaria; Levi, Sonia; Volonte, Giovanna; Paini, Cinzia; De Lalla, Claudia; Siccardi, Antonio G.; Baralle, Francisco E.; et al.
CORPORATE SOURCE: Dip. Ric. Biol. Tecnol., Ist. Sci. H San Raffaele, Milan, 20132, Italy
SOURCE: Journal of Biological Chemistry (1993), 268(29), 21819-25
CODEN: JBCHA3; ISSN: 0021-9258
DOCUMENT TYPE: Journal
LANGUAGE: English

Search completed by David Schreiber 308-4292

AB The mol. cloning of the cDNA encoding for an isoallergenic form of Lol pII, a major rye **grass** (Lolium perenne) pollen allergen, was performed by PCR amplification on mRNA extd. from pollen. The amino acid sequence derived from the cDNA was truncated by 4 and 5 residues at the N- and C-terminal ends, resp., and differenced only in one position from that previously reported. This cDNA was expressed in Escherichia coli by fusion to the C-terminus of the human ferritin H-chain. The mol. was produced in high yields as a sol. protein and was easily purified. The protein retains the multimeric quaternary structure of ferritin, and it exposes on the surface the allergenic moiety, which can be recognized in Western blotting and in ELISA expts. by specific IgE from allergic patients. The recombinant allergen was used to analyze the sera of 26 patients allergic to L. perenne compared with control sera. The results were in good agreement with the values obtained with the radioallergosorbent test assay. In addn., histamine release expts. in whole blood from an allergic patient and skin prick tests showed that the recombinant allergen retains some of the biol. properties of the natural compd. These findings indicate that the availability of homogeneous recombinant allergens may be used for the development of more specific diagnostic and therapeutic procedures. Moreover, this expression system may be of more general interest for producing large amts. of sol. protein domains in E. coli.

L24 ANSWER 27 OF 36 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V. on STN
ACCESSION NUMBER: 93270757 EMBASE
DOCUMENT NUMBER: 1993270757
TITLE: Purification and characterization of recombinant Bet v I, the major birch pollen **allergen**. Immunological equivalence to natural Bet v I.
AUTHOR: Ferreira F.D.; Hoffmann-Sommergruber K.; Breiteneder H.; Pettenburger K.; Ebner C.; Sommergruber W.; Steiner R.; Bohle B.; Sperr W.R.; Valent P.; Kungl A.J.; Breitenbach M.; **Kraft D.**; Scheiner O.
CORPORATE SOURCE: IAEP, Universitat Wien, Wahringer Gurtel 18-20,A-1090 Vienna, Austria
SOURCE: Journal of Biological Chemistry, (1993) 268/26 (19574-19580).
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Pollen from trees of the order Fagales (e.g. birch, alder, hazel, oak, and hornbeam) are a major cause of Type I allergies observed in early spring. Previously, we reported the cloning and sequencing of Bet v I, the major birch pollen **allergen**, which showed high sequence similarities to a family of plant pathogen-activated genes (Breiteneder, H., Pettenburger, K., Bito, A., Valenta, R., Kraft, D., Rumpold, H., Scheiner, O., and Breitenbach, M. (1989) EMBO J. 8, 1935-1938). Here, we present the results on the expression, purification, and characterization of recombinant Bet v I produced in Escherichia coli as fusion and non-fusion protein, respectively. The purified recombinant proteins were analyzed to verify purity and structural integrity, and their immunological properties were compared to those of Bet v I isolated from birch pollen (natural Bet v I). Immunoblot analyses showed that the recombinant proteins are specifically recognized by monoclonal antibodies raised against natural Bet v I as well as by IgE from birch pollen-allergic patients. However,

enzyme-linked immunosorbent assays revealed a decreased IgE-binding activity of the recombinant fusion Bet v I compared to the non- fusion and natural Bet v I proteins, which probably results from conformational changes due to the fusion tail. Recombinant non-fusion Bet v I was equivalent to natural Bet v I with respect to IgE-binding properties, the ability to induce in vitro proliferation of **allergen**-specific T-cell clones, and the ability to release histamine from basophils derived from birch pollen-allergic patients.

L24 ANSWER 28 OF 36 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
ACCESSION NUMBER: 93:459223 SCISEARCH
THE GENUINE ARTICLE: LN986
TITLE: ANTIGEN-SPECIFIC AND ISOTYPE-SPECIFIC IMMUNE-RESPONSES TO A RECOMBINANT ANTIGEN-**ALLERGEN CHIMERIC** (RAAC) PROTEIN
AUTHOR: ZHANG L; MOHAPATRA S S (Reprint)
CORPORATE SOURCE: UNIV MANITOBA, DEPT IMMUNOL, 730 WILLIAM AVE, WINNIPEG R3E 0W3, MANITOBA, CANADA
COUNTRY OF AUTHOR: CANADA
SOURCE: JOURNAL OF IMMUNOLOGY, (15 JUL 1993) Vol. 151, No. 2, pp. 791-799.
ISSN: 0022-1767.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 41

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Ag-specific IgE and IgG antibody responses to a recombinant Ag-**allergen chimeric** (RAAC) protein were examined in B6D2F1 mice. The RAAC protein consisted of the truncated beta-galactosidase (beta-gal), linked at its C terminus to a polypeptide representing the conserved region of the recombinant Kentucky Bluegrass allergen encoded by the cDNA clone KBG8.3(rKBG8.3). Immunization of the mice with the RAAC protein in dextran sulfate as adjuvant led to the differential production of antibodies to the two constituents of RAAC protein with respect to their isotypic classes. Most of the antibody responses to both sets of determinants on the fusion protein were IgG1. In addition, the allergenic polypeptide of RAAC protein induced IgE antibodies, whereas the beta-gal elicited IgG2a antibodies. The same pattern of antibody isotypes was produced when the individual components, rKBG8.3 and beta-gal were separately used for immunization with dextran sulfate. On the other hand, immunization of mice with either RAAC or the beta-gal in CFA induced primarily a IgG response, and no IgE antibodies; however, under the same conditions of immunization rKBG8.3 induced IgG1 antibodies and also low levels of IgE antibodies. In contrast, the RAAC and the beta-gal induced IgG2a antibodies, whereas rKBG8.3 induced no detectable IgG2a antibodies. Furthermore, high titers of IgE antibodies were induced by the rKBG8.3 and not by the RAAC protein in dextran sulfate after the mice had been immunized twice with the same polypeptide in CFA. It is inferred from these results that the induction of isotype-specific immune responses in the animals with the same genetic background is dependent upon the Ag in question as well as the adjuvant; the latter, however, influences the magnitude but does not determine the isotype of the immune responses.

L24 ANSWER 29 OF 36 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
ACCESSION NUMBER: 93:719904 SCISEARCH
THE GENUINE ARTICLE: MJ388
TITLE: MOLECULAR AND FUNCTIONAL-CHARACTERIZATION OF ALLERGENS - BASIC AND PRACTICAL ASPECTS

AUTHOR: SCHEINER O (Reprint)
CORPORATE SOURCE: UNIV VIENNA, INST ALLGEMEINE & EXPTL PATHOL, WAHRINGER GURTEL 18-20, A-1090 VIENNA, AUSTRIA (Reprint)
COUNTRY OF AUTHOR: AUSTRIA
SOURCE: WIENER KLINISCHE WOCHENSCHRIFT, (1993) Vol. 105, No. 22, pp. 653-658.
ISSN: 0043-5325.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: CLIN
LANGUAGE: German
REFERENCE COUNT: 64

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Well-defined allergens are a prerequisite for the exact diagnosis and immunotherapy of Type I (IgE-mediated) allergic diseases. The allergens have to be available in highly purified form and in sufficient quantities. By applying molecular cloning methods this goal can be achieved with respect to both characterization and reproducibility of allergen preparations. Moreover, this technique leads to the deduction of primary structures of allergens, which allows computer-aided comparisons with already known amino acid sequences. Significant sequence similarities with well-described proteins may point to a biological and biochemical function of the cloned allergen. The major allergen of white birch, Bet v 1, and its close relatives from alder, hazel, and hornbeam belong to a family of pathogenesis-related proteins ubiquitous in angiosperms. Based on these results, a percentage of food intolerance can now be regarded as Type I allergy due to Bet v 1-related proteins. By sequence comparisons, Bet v 2, another birch pollen allergen, was identified as the ubiquitous cytoskeleton-associated protein, profilin. For its high sequence conservation and its ubiquitous appearance in allergenic sources of plant origin, profilin was shown to represent a pan-**allergen**. Recombinant non-**fusion** Bet v 1 revealed identical immunological properties with respect to interaction with both antibodies and Bet v 1-specific T cell clones when compared with natural Bet v 1 purified from birch pollen.

L24 ANSWER 30 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1993:122996 HCAPLUS
DOCUMENT NUMBER: 118:122996
TITLE: Protein allergens of Cynodon dactylon, their recombinant production, nucleic acid sequences, and diagnostic and therapeutic uses
INVENTOR(S): Knox, Robert Bruce; Singh, Mohan Bir; Smith, Penelope Mary
PATENT ASSIGNEE(S): University of Melbourne, Australia
SOURCE: PCT Int. Appl., 52 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9216554	A1	19921001	WO 1992-AU108	19920313
W: AU, JP, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, MC, NL, SE				
AU 9215662	A1	19921021	AU 1992-15662	19920313
PRIORITY APPLN. INFO.:			AU 1991-5084	19910314
			WO 1992-AU108	19920313

Search completed by David Schreiber 308-4292

AB Nucleic acid sequences coding for protein allergens of Cynodon dactylon (Bermuda **grass**) pollen are presented which are cloned and expressed in host cells transformed with the nucleic acid. The isolated protein allergens or allergenic fragments are useful for diagnosing and treating individuals sensitive to Bermuda **grass** pollen allergens. Three cDNA clones encoding Bermuda **grass** pollen allergens were isolated and characterized and produced as fusion proteins with .beta.-galactosidase or glutathione-S-transferase. Nucleotide sequences for the clones are shown. Nine of 12 sera of patients with allergy to Bermuda **grass** reacted with clones B1 and B4 as fusion proteins; 6 of 12 reacted with clone B2 as fusion protein.

L24 ANSWER 31 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1992:569421 HCAPLUS
DOCUMENT NUMBER: 117:169421
TITLE: Birch pollen allergen P14 for diagnosis and therapy of allergic diseases, and recombinant production of the allergen
INVENTOR(S): Valenta, Rudolf; Duchene, Michael; Pettenburger, Karin; Breitenbach, Michael; Kraft, Dietrich; Rumpold, Helmut; Scheiner, Otto
PATENT ASSIGNEE(S): Biomay Biotechnik Produktions- und Handelsgesellschaft m.b.H., Austria
SOURCE: PCT Int. Appl., 69 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9203551	A1	19920305	WO 1991-EP1513	19910809
W: AU, CA, FI, JP, NO, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE				
AT 9001685	A	19951115	AT 1990-1685	19900813
AT 401180	B	19960725		
CA 2067182	AA	19920214	CA 1991-2067182	19910809
AU 9183901	A1	19920317	AU 1991-83901	19910809
AU 659609	B2	19950525		
EP 495064	A1	19920722	EP 1991-914581	19910809
EP 495064	B1	20010711		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
JP 05502589	T2	19930513	JP 1991-513486	19910809
AT 203055	E	20010715	AT 1991-914581	19910809
NO 9201375	A	19920612	NO 1992-1375	19920408
FI 9201600	A	19920410	FI 1992-1600	19920410
US 5583046	A	19961210	US 1992-846992	19920606
US 5648242	A	19970715	US 1995-469555	19950606

PRIORITY APPLN. INFO.:

AT 1990-1685	A	19900813
US 1991-683832	A	19910411
WO 1991-EP1513	A	19910809
US 1992-846992	A3	19920606

AB Recombinant DNAs are provided which code for polypeptides having the antigenicity of P14 allergen of birch (Betula verrucosa) and other plants of the order Fagales (and for polypeptides comprising .gtoreq.1 epitope thereof). Methods of producing the proteins and polypeptides are disclosed, as is their use in diagnosis and therapy of allergic diseases. A method for purifn. of P14 allergens or cross-reactive allergens using

binding to poly(L-proline) is also disclosed. The polynucleotide coding for birch P14 was inserted in plasmid pKK223-3 for prodn. of a recombinant nonfusion protein, while a recombinant fusion protein was produced using plasmid pEXB. Reactivity of the produced polypeptides with patient IgE is shown. Homol. of the birch P14 sequence with a variety of profilin sequences is included.

L24 ANSWER 32 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1993:121184 HCAPLUS
DOCUMENT NUMBER: 118:121184
TITLE: The genes for allergens of Kentucky bluegrass and the manufacture of the allergens in heterologous hosts
INVENTOR(S): Mohapatra, Shyam S.; Hill, Robert D.; Kisil, Fred T.; Sehon, Alec
PATENT ASSIGNEE(S): Can.
SOURCE: Can. Pat. Appl., 55 pp.
CODEN: CPXXEB
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
CA 2066801	AA	19921027	CA 1992-2066801	19920422
PRIORITY APPLN. INFO.:			US 1991-693236	19910426
			US 1991-726937	19910708

AB Novel genes for allergens of Kentucky bluegrass (Poa pratensis) pollen are cloned and sequenced and the proteins manufd. in an heterologous host for use in the diagnosis and treatment of allergies and asthma. A P. pratensis pollen cDNA library in .lambda.gt11 was screened with sera from patients allergic to the pollen. Three clones were found; their sequences were very similar to one another but showed only sporadic similarity to other sequences; sequences cross-hybridizing with these sequences and material cross-reacting with antibody to the proteins was found in pollen of several other species. These sequences appear to be members of a multigene family that is expressed in pollen but not in leaf. The cDNA for one of these allergens was expressed in Escherichia coli using expression vectors of the prior art. IgG-binding epitopes of the proteins were identified.

L24 ANSWER 33 OF 36 HCAPLUS COPYRIGHT 2003 ACS on STN
ACCESSION NUMBER: 1993:99865 HCAPLUS
DOCUMENT NUMBER: 118:99865
TITLE: Expression and thrombin cleavage of Poa p IX recombinant allergens fused to glutathione S-transferase
AUTHOR(S): Olsen, Egil; Mohapatra, Shyam S.
CORPORATE SOURCE: Dep. Immunol., Univ. Manitoba, Winnipeg, MB, R3E OW3, Can.
SOURCE: International Archives of Allergy and Immunology (1992), 98(4), 343-8
CODEN: IAAIEG; ISSN: 1018-2438
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The high-level expression and purifn. of Poa p IX recombinant **grass** pollen allergens were examd. utilizing a modified pGEX plasmid, designated as pGEX 2T-1. This vector permits frame-1 ligation of .lambda.gt11 cDNA inserts and cleavage of the recombinant allergenic

protein from the fusion partner glutathione S-transferase. The expression of the fusion proteins in water-sol. form varied among the transformants of the same bacterial strain and also between different host strains. Purifn. of the fusion proteins by affinity chromatog. employing glutathione agarose gel revealed that proteases in the bacterial lysate bound to the gel and were co-eluted with the fusion proteins. These proteases, which specifically degraded the recombinant proteins to varying degrees, were inhibited by both of the inhibitors, PMSF and aprotinin. Cleavage by thrombin of the fusion proteins indicated that the structure of the individual protein affected the thrombin accessibility to the cleavage site. Increased concn. of thrombin partly compensated this effect, but resulted in a broader specificity of the enzyme. By contrast, cleavage of the fusion protein when it was still attached to the glutathione gel was convenient and led to purifn. of the product devoid of proteolytic activity. Since almost all the recombinant allergens have been cloned in .lambda.gt11 vector, the pGEX 2T-1 vector reported herein will facilitate the synthesis, purifn. of the corresponding allergenic proteins or their peptides in sol. and biol. active forms.

L24 ANSWER 34 OF 36 MEDLINE on STN DUPLICATE 8
ACCESSION NUMBER: 92283560 MEDLINE
DOCUMENT NUMBER: 92283560 PubMed ID: 1597349
TITLE: Diagnosis of **grass** pollen allergy with recombinant timothy **grass** (Phleum pratense) pollen allergens.
AUTHOR: Valenta R; Vrtala S; Ebner C; Kraft D; Scheiner O
CORPORATE SOURCE: Institute of General and Experimental Pathology, AKH, University of Vienna, Austria.
SOURCE: INTERNATIONAL ARCHIVES OF ALLERGY AND IMMUNOLOGY, (1992) 97 (4) 287-94.
Journal code: 9211652. ISSN: 1018-2438.
PUB. COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199207
ENTRY DATE: Entered STN: 19920717
Last Updated on STN: 19920717
Entered Medline: 19920707

AB In order to establish a test system for **grass** pollen allergy based on the use of recombinant allergens we chose timothy **grass** (Phleum pratense), a widely spread **grass**, as a model. From a lambda gt11 cDNA expression library that we had constructed from pollen RNA of timothy **grass** (P. pratense), we had obtained with serum IgE from a **grass** pollen-allergic individual 60 IgE-binding clones. By differential testing with sera from different **grass** pollen-allergic patients, we selected three distinct clones encoding **Phl p I** (group **I**), **Phl p** V (group V) and profilin from timothy **grass**, which when used together allowed the diagnosis of **grass** pollen allergy in 97 out of 98 tested **grass** pollen-allergic patients employing a simple plaque lift technique. This recombinant test based on plaque lifts containing **allergen**-beta-galactosidase **fusion** proteins was compared with IgE immunoblots using crude pollen protein extracts from timothy **grass**. Both methods were in good agreement with RAST scores and clinical data, and proofed to be useful for the diagnosis of **grass** pollen allergy. Our results further indicate that a limited panel of only two recombinant **grass** pollen allergens, **Phl p I** and Phl p V,

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together with the plant panallergen profilin could be sufficient for the diagnosis and possibly immunotherapy of **grass** pollen allergy.

L24 ANSWER 35 OF 36 MEDLINE on STN DUPLICATE 9
ACCESSION NUMBER: 91007960 MEDLINE
DOCUMENT NUMBER: 91007960 PubMed ID: 2210872
TITLE: Isolation and characterization of a cDNA clone encoding an IgE-binding protein from Kentucky bluegrass (Poa pratensis) pollen.
AUTHOR: Mohapatra S S; Hill R; Astwood J; Ekramoddoullah A K; Olsen E; Silvanovitch A; Hatton T; Kisil F T; Sehon A H
CORPORATE SOURCE: Department of Immunology, Medical Research Council Group for Allergy Research, University of Manitoba, Winnipeg, Canada.
SOURCE: INTERNATIONAL ARCHIVES OF ALLERGY AND APPLIED IMMUNOLOGY, (1990) 91 (4) 362-8.
Journal code: 0404561. ISSN: 0020-5915.
PUB. COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199011
ENTRY DATE: Entered STN: 19910117
Last Updated on STN: 19910117
Entered Medline: 19901120

AB We reported previously on the isolation and characterization of several allergens from Kentucky bluegrass (KBG) (Poa pratensis L.) pollen with the aid of the corresponding murine monoclonal antibodies (Mabs). In the present study, (1) an analysis of various tissues of this **grass** revealed that the allergenic components recognized by these Mabs were confined to the pollen; (2) intact translatable mRNA was isolated from the KBG pollen, and (3) a cDNA library was constructed with this mRNA in the lambda gt11 expression vector. Screening of this library with a pool of six sera from KBG-allergic patients, in combination with enzyme-labeled antibodies to human IgE, led to the isolation of a cDNA clone, referred to as KBG7.2. The nick-translated cDNA probe of KBG7.2 hybridized to a 1.5-kbp RNA transcript from KBG pollen. Moreover, transcripts corresponding to KBG7.2 were found in pollens of eight other **grasses**, indicating that the proteins similar to the one encoded by this cDNA may be present in these **grasses**. The nucleotide sequence of KBG7.2 was determined; interestingly, the corresponding derived amino acid sequence did not match any other sequence recorded in the protein data banks. The peptide encoded by KBG7.2 was expressed as a fusion protein utilizing the plasmid vector pWR590.1. Whereas none of the above **allergen**-specific Mabs bound to the **fusion** protein, all the 15 individual sera from **grass** pollen allergic patients recognized the fusion protein.(ABSTRACT TRUNCATED AT 250 WORDS)

L24 ANSWER 36 OF 36 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN DUPLICATE 10
ACCESSION NUMBER: 1990:43698 BIOSIS
DOCUMENT NUMBER: BA89:21062
TITLE: CLONING OF COMPLEMENTARY DNA CODING FOR AN ALLERGEN OF COCKSFOOT **GRASS** DACTYLIS-GLOMERATA POLLEN.
AUTHOR(S): WALSH D J; MATTHEWS J A; DENMEADE R; WALKER M R
CORPORATE SOURCE: DEP. CLIN. CHEM., UNIV. BIRMINGHAM, WOLFSON RES. LAB., QUEEN ELIZABETH MED. CENT., EDGBASTON, BIRMINGHAM B15 2TH, UK.
SOURCE: INT ARCH ALLERGY APPL IMMUNOL, (1989) 90 (1), 78-83.

Search completed by David Schreiber 308-4292

CODEN: IAAAAM. ISSN: 0020-5915.
FILE SEGMENT: BA; OLD
LANGUAGE: English

AB Messenger RNA isolated from Cocksfoot **grass** (Dactylis glomerata) anthers has been used to generate a cDNA library in .lambda.t11. Three cDNA clones (7.8, 8.1, and 8.3) were demonstrated to be recognized by human IgE antibodies in atopic serum and by rabbit polyclonal antiserum raised to a crude aqueous extract of Cocksfoot pollen. The size of the cDNA inserts was determined as approximately 700 bp, and restriction mapping demonstrated them to be identical sequences. Lysogens obtained in Escherichia coli Y1089 allowed expression of a 140 kD .beta.-galactosidase fusion protein containing 24 kD of cloned **allergen** protein. **Fusion** proteins were recognized by IgE antibodies in 75% (6/8) of atopic sera tested, but were not detected by non-atopic sera. On the basis of size and frequency of recognition in the atopic population, the cloned protein may represent a major allergen. Monoclonal antibodies specific for the major allergen of Cocksfoot pollen were not reactive with the fusion proteins. Reactivity of human IgE antibodies with the fusion protein could be blocked by crude Cocksfoot pollen extract, but not by the major allergen DG3 purified from the extract by affinity chromatography. Human and rabbit antibodies affinity purified against fusion protein 7.8 did not allow identification of the native protein component in crude extract encoded for by the cDNA clones.